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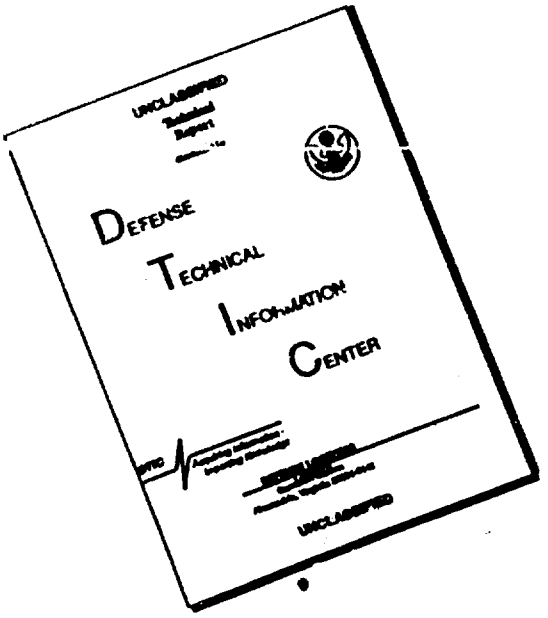

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Action of Infrared Rays on Microorganisms, by G. D. Goncharov

The staging of tests on the action of infra-red rays (IRR) on a live organism depends on the understanding of the very nature of the IRR. If we have in mind the electromagnetic theory of light, that is, that the IRR, while causing an oscillatory movement of high frequency of the small particles of a substance encountered in their path, render an exceptionally thermal energy by this reason which raises the temperature of this substance, than the action of the IRR on the organism can be exclusively considered as an action of a thermic factor which stimulates or hinders the vital activity of the cell or kills the cell, depending on the intencity of the heating. Then the tests should be run so that the object being irradiated is completely isolated from convectional warmth which is escaping from the source of the IRR, considering only the radiant heat.

If we consider that the electromagnetic oscillation renders an influence to the cell, and along with this an increase in the temperature of the cell, than we should demarcate the action of both factors; thermic and electromagnetic oscillation of a determined frequency. Then the tests should be run so that the objected being irradiated is fully isolated from the total action of the convectional and ray heat, that is, from the basic thermic properties of the IRR, considering only the action of the electromagnetic oscillation. The test run by us meet these requirements.

In 1907, when there were already numerous studies (Arloing, Ward, Bie) which noted the bactericidal properties of the IRR, Wiesner pointed out that the IRR possess significant bactericidal actions (on Staphylococcus pyogenes aureus) and

at the same time confirming the first observations of Geisler in the smoky (sooty) retorts with typhus baccilli. Wiesner utilized cast iron plates as the source of the IRR. They were heated on a burner with special adaptors which allowed regulation of the intencity of the (e) radiation and avertion of the convectional transmission of the heat to the object being irradiated. The control object is put into a themostat at a temperature equal to the temperature of the air of the testing enclosure. The temperature in the testing enclosure is never increased higher than the temperature which is menacing to the life of the bacteria. Wiesner also stresses that it is necessary that there be no alteration of agar plate on which the colonies of bacteria are growing during this temperature under irradiation; the temperature of the plate should be higher than the temperature of the surrounding air (because it does not cool) which, evidently, stipulates the lethal action of the IRR. Further on in his tests on the effect of the temperature of the air during irradiation, Wiesner was convinced that during the same intencity of (e) radiation, but during an increase of the temperature of the air, the bactericidal action of the rays increased, and during lowering of the temperature the action of them decreased. Irregardless of this, Wiesner comes to the conclusion that the bactericidal action of the IRR is not a thermic effect.

Work of recent years (Nazaroba, Glykson, Arnautov) which speak in favour of the specific action of the IRR do not differ with any persuasiveness of data because of the lack of physical conditions in the tests in which the action of the IRR was not expressed, but the total actions of the convectional and radiant heat and, besides this, the said authors, utilizing an electric stove without a filter as the source of IRR, were working not with IRR alone but with other rays

which adjoined the tests.

In our first tests, as was reported earlier (Goncharov 1937), the object being irradiated was not sufficiently isolated from the convectional heat. Therefore we had a tendency to contend the presence of thermic action of the IRR exclusively. Later we built a cryostat for the ellimination of the convectional heat, this was a two phase metal box with a movable screening partition-shelf. On one side the box has a cut-out 15 by 15 cm on which is secured a filter with a cooler. The vessel with the object being irradiated is put into the box on the cooling support. All the cooling of the cryostat, as will as the filter and support, was done with flowing water.

To elliminate the possibility of the heat factor, control objects were placed into a thermostat with the same temperature that was present during the irradiation, the medium before sowing was subjected to a thermostat until the disired temperature was attained to avert an effect of fast heating during comparison of the conttrol and testing objects. The temperature of the medium being irradiated was always under control in contrast to the tests of Wiesner. The controling was done with the aid of a thermocouple according to the difference the deviations of the mirror-galvanometer.

The source of the IRR was a 1000 W. electric lamp operating on 110 v. The distance from the lamp filament to the surface of the medium with the culture was as near as possible to 19 cm in order to obtain the best effect. A filter released the rays, wave lenght 0.8 to 1.2 microns, that is, rays immediately adjoining to the visible red rays.

Because the tests were mainly with a fluid nutritive medium it was necessary to establish to what extent the IRR were retained by the medium. With the aid

of a thermotrunk it was established that at a distance of 19 cm from the lamp filament through a 15 mm layer of the nutritive medium in Kokca vessels there is the following from the general stream of the IRR; through liquid in 8,5 B11--9.5%; through Ghiltan medium--12.4%.

Four other series of tests were run. Irradiation was conducted in 100 cm3 of the wash B11, 15 mm high in a Kokha vessel, there was added 1 cm3 of a yeast suspension of a 2 day agar culture. Determination of the possible fermentative variations after the irradiation was done by determining the quantity of sugar in the liquid in the process of alcohal fermentation according to Bertran. Determinations were made every 24 hours. The cultures were irradiated 4 hours each day for 3 days. The temperature of the wash during irradiation was near 37-42 C.

The quantity of maltose remaining at the end of the main fermentation in the irradiated and non-irradiated cultures was percentage wise 0.1, which is in the levels of a possible error. The weight of sediment determined at the end of the fermentation process, having been dried in an desiccator, also fluctuated in the levels of 0.1%.

Under these conditions of the test the IRR did not render any effect on the fermentative properties of the yeast to ferment the maltose. Morphological variations of the yeast cells during this were not noted either.

The bacteria Bact. prodigiosum and Pseudomonas fluorescens liquefaciens were used in the following tests.

The Bact. prodigiosum was subjected to irradiation with a 4 hour exposure. There were three tests. The temperature of the medium during the irradiation was maintained near 46-47 C. One control culture was at this temperature and for this lenght of time, that is, 4 hours, in a therrostat (wrapped in black photo-wrapping

paper); the other control was at 30 C. in a thermostat- the temperature at which all the cultures were held after the test. The results of the tests indicate that the action of the IMR tended to hinder the formation of colorless colonies (leukoraces); a part of the colonies on the irradiated vessels remained red while the control at this same temperature were all colorless.

Analogical tests were conducted with Ps. fluorescens liquefaciens on 'MPA' and on an agar medium, Giss, with glucose. Variations of the thermic nature were noted here also, and in particular on the Giss medium because the temperature of the irradiation medium and control equaled 45-46 C. These variations were fixed by photo-survey of seperate dailey colonies, resulting after irradiation and after the action of the temperature at 45-46 C in a thermostat. The variations were: un-linking of the original form into two types of colonies N and R in the vessels subjected to irradiation as well as those in the thermostat (exposure- 2 hours; thermostat temperature-45-46 C.). The splitting action was not noted at 30 C. The colonies were of one type.

During a 4 hour irradiation at 45 C. in a thermostat there were only growths of transparent, diffused colonies of the type N. At 35 C. in a thermostat colonies of the original type grew out, as also at 30 C., with two concentric zones sharply broken in the center.

Successive tests with Ps. fluorescens liquefaciens were conducted on the Chilton medium with a count of their denitrification ability in forming nitrites. Determination of the nitrites was done colorimetrically according to Dubosc with iodine-starch-zinc. The results of the colorimetrical observations are in Table 1. The method of irradiation used is the same as that on Saccharomyces cerevisiae in a fluid medium. Utilizing the possibility of regulating the temperature (in

levels of 2 C) of the culture being irradiated in the cryostate, the tests were conducted at various temperatures with simultaneous control in the thermostate of this same temperature.

Numerous test indicated that the maximal speed of the process of denitrification of the culture being irradiated is at 27-29 C. and at 41-42 C. As Table one shows, the process of denitrigication of the culture always is faster in the irradiated cultures: at 46-47 C.-- on the 3rd day for the irradiated culture and on the 4th for the control; at 41-42- irradiated 2nd day, control 4th day; at 27-29 C.- irradiated 2nd day and control 4th day; at 15-17 C. in the irradiated 4th day and control 5th day. Further observations on the gas change in the conditions of IRR would undoubtebly give us the answer as to why there is a speeding up of the denitrification in the cultures on the Chilton medium.

Along with this, as was expected, there was observed that the increase of the temperature in the cultures caused a variation in the size of the bacilli and the appearence of vacuolation. Some of the most characteristic moments were pictured by microphotographic means. The size of the irradiated and control culture baccilli at an equal temperature was somewhat different. The irradiated culture baccilli were from 1.8 to 3.0 microns; in the control culture at this same temperature- from 1.5 to 2.8 microns, and sharply differed from the control at 30 C. where the baccilli were from 0.9 to 2.2 microns with the predominant size 0.9 microns. Thus an increase of the temperature slows the process of division in the irradiated cultures also.

Conclusions

Although our work had a preliminary character, according to the conditions of the said tests, which allowed for the isolation of the object being irradiated

from the sum action of the convectional and radiated heat, we can conclude that the action of the IRR, having a wave lenght of 0.8-1.2 microns:

1. did not reflect on the fermentative properties of the Saccharomyces cerevisiae XII in the fermenting of the maltose and did not reflect on the morphology of the yeast cells;

2. tended to hinder the formation of the leukoraces of the Bact. prodigiosum at 46-47C.;

3. did not reflect on the character of the structure of the colonies of the Pseudomonas fluorescens liquefaciens;

4. stimulated the process of denitrification in the culture Pseudomonas fluorescens liquefaciens on the Chiltan medium;

5. slighly hindered the process of division in regard to the lenghtening of the cells.

In closing I express my deep appreciation to Prof. P. L. Isachenko for a numerous valuable aids in this work and also Prof. G. M. Frank.

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Table 1. Denitrification ability of Pseudomonas fluorescens liquefaciens (NO_2 in mg on 1.5 cm3)

Time of determination of nitrite	Temperature 46-47		41-42		27-29		25-27	
	Irradiation	Thermostat	Irradiation	Thermostat	Irradiation	Thermostat	Irradiation	Thermostat
On 2nd day prior to 2nd action of IRR and tempera.	0,0382	0,0428	0,0334	-	0,0597	0,0537	0,0443	0,0463
2nd day after 2nd action of IRR and temperature.	0,0417	0,0423	0,00	0,0262	0,00	0,0597	0,0569	0,0555
3rd day after 2nd action of IRR and tempatature.	0,00	0,0417	-	0,0252	-	++++	0,0492	0,0368
4th day after.	-	0,00	-	0,00	-	0,00	0,00	0,0154
5th day after.	-	-	-	-	-	-	-	0,00

- No nitrite: ++++ Qualitive determination of a very large quantity of nitrite